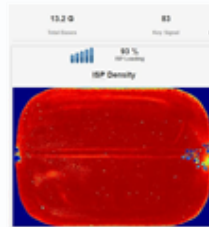
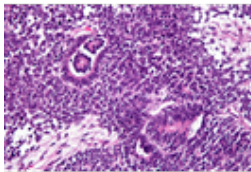


Clinical Next Generation Sequencing

Introducing Oncology Hotspot

The Oncology Hotspot assay from Mount Sinai Genetic Testing Laboratory is intended to detect clinically actionable genetic alterations in patients with advanced solid tumor cancer, typically identified as Stage 2 or 3 or metastatic (Stage 4) cancer. Our Oncology Hotspot assay is available for use with any advanced solid tumor patient. In particular, if you are treating advanced melanoma, lung or colorectal cancer patients, our oncology hotspot assay will provide actionable insights for BRAF variants; specific EGFR variants; and KRAS, BRAF, PIK3CA, and AKT mutant versus wild type reporting as clinically indicated, in addition to any other clinically relevant variants detected in the remaining genes.

Using a combination of multiplex PCR and next generation sequencing (NGS) technologies, our Oncology Hotspot assay analyzes 207 regions of the genome to determine if any variants identified are known drivers of cancer and whether any drug response or resistance is associated with the variant(s), providing you with a clear and insightful set of information as part of your overall treatment and care management plan. Hotspot assays, separate from whole exome or whole genome sequencing, look for specific variants within specific regions of the genes being tested versus detecting variants in the entire coding sequence of each gene.



Specimen requirements

The desired specimen for this assay is:

- Nine (9) unstained glass slides
- One (1) H&E stained glass slide

While the Oncology Hotspot assay is designed to detect variants down to 5% allele fraction, we require a tumor specimen submission to contain at least 20% tumor cells to account for tumor heterogeneity.

Results reporting and Turnaround Time (TAT)

The identified genetic alterations receive expert review and clinical utility interpretation by our molecular pathologists and geneticists. We will provide a faxed result report upon completion of the testing and have our board certified pathologists and geneticists available by phone to answer any questions you may have. We continue to optimize our testing procedures to ensure as fast a turnaround time as possible, anticipating 10 – 21 days from the accessioning of the sample to the result report.

Detailed gene list for the Hotspot Oncology assay

The Oncology Hotspot assay looks for hotspot variants in 50 genes that are associated with cancer diagnosis, prognosis, and response: *ABL1, AKT1, ALK, APC, ATM, BRAF, CDH1, CDKN2A, CSF1R, CTNNA1, EGFR, ERBB2, ERBB4, EZH2, FBXW7, FGFR1, FGFR2, FGFR3, FLT3, GNA11, GNAQ, GNAS, HNF1A, HRAS, IDH1, IDH2, JAK2, JAK3, KDR, KIT, KRAS, MET, MLH1, MPL, NOTCH1, NPM1, NRAS, PDGFRA, PIK3CA, PTEN, PTPN11, RB1, RET, SMAD4, SMARCB1, SMO, SRC, STK11, TP53, VHL.*

| Gene | % Mutated in all Tumors | Specific Tumor Type | Clinical Implications of Variants |
|----------------|-------------------------|---|-------------------------------------|
| <i>ABL1</i> | 5 | | Clinical Studies |
| <i>AKT1</i> | 1 | | EGFR resistance; Clinical Studies |
| <i>AL</i> | 3 | Neuroblastoma | ALK inhibitors (1) |
| <i>APC</i> | 7 | Colorectal 40% | Clinical Studies |
| <i>ATM</i> | 4 | Colorectal 9%; Skin 8% | Clinical Studies |
| <i>BRAF</i> | 20 | Melanoma 50%; Thyroid 50%; Colorectal 10% | Response to RAF inhibitors |
| <i>CDH1</i> | 2 | Breast 11%; Stomach 11% | Diagnostic for Diffuse/Lobular type |
| <i>CDKN2A</i> | 8 | Skin 18% | Clinical Studies |
| <i>CSF1R</i> | 1 | | Clinical Studies |
| <i>CTNNB1</i> | 8 | Soft tissue 40% | Clinical Studies |
| <i>EGFR</i> | 20 | Lung 15% | Response and Resistance to EGF |
| <i>ERBB2</i> | 1 | Small intestine 10% | HER pathway inhibitors |
| <i>ERBB4</i> | 2 | Skin 15% | HER pathway inhibitors (2) |
| <i>EZH2</i> | 2 | | Clinical Studies |
| <i>FBXW7</i> | 3 | Uterine 9%; Colorectal 9% | Clinical Studies |
| <i>FGFR1</i> | 1 | | Clinical Studies |
| <i>FGFR2</i> | 1 | Uterine 8% | Clinical Studies |
| <i>FGFR3</i> | 8 | Urinary Tract 35% | Clinical Studies |
| <i>FLT3</i> | 15 | Hemeatopoietic 20% | Clinical Studies |
| <i>GNA11</i> | 1 | Eye 37% | Diagnostic for uveal melanoma |
| <i>GNAQ</i> | 2 | Eye 37% | Diagnostic for uveal melanoma |
| <i>GNAS</i> | 3 | Pseudomyxoma 71%; Pituitary 30%; Pancreas 14% | Diagnostic |
| <i>HNF1A</i> | 1 | Liver 7% | Clinical Studies |
| <i>HRAS</i> | 2 | Salivary Gland 12%; Skin 11% | Clinical Studies |
| <i>IDH1</i> | 11 | CNS 35% | Good prognosis in CNS |
| <i>IDH2</i> | 3 | CNS 1.5% | Good prognosis in CNS |
| <i>JAK2</i> | 31 | Common in myeloproliferative disorders | Clinical Studies |
| <i>JAK3</i> | 1 | | Clinical Studies |
| <i>KDR</i> | 2 | | Clinical Studies |
| <i>KIT</i> | 12 | Up to 85% of GIST | Kinase inhibitors |
| <i>KRAS</i> | 15 | Pancreatic 92%; Colorectal 40% | Resistance to EGFR inhibitors |
| <i>MET</i> | 2 | Kidney 3% | Clinical Studies |
| <i>MLH1</i> | 1 | Colorectal 4% | Microsatellite instability |
| <i>MPL</i> | 2 | Clinical Studies | |
| <i>NOTCH1</i> | 6 | Head and Neck 13%; Skin 9% | Clinical Studies |
| <i>NPM1</i> | 10 | Common in AML | Clinical Studies |
| <i>NRAS</i> | 5 | Skin 15%; Thyroid 7% | Resistance to EGFR inhibitors |
| <i>PDGFRA</i> | 4 | Up to 25% GIST | Kinase inhibitors |
| <i>PIK3CA</i> | 15 | Breast 25%; Colon 20% | Resistance to EGFR inhibitors |
| <i>PTEN</i> | 6 | Endometrial 39%; CNS 13%; Skin 9%; Breast 7% | Clinical Studies |
| <i>PTPN11</i> | 2 | | Clinical Studies |
| <i>RB1</i> | 3 | | Diagnostic for retinoblastoma |
| <i>RET</i> | 3 | Thyroid 25% | RET inhibitors |
| <i>SMAD4</i> | 3 | GI 50% | Clinical Studies |
| <i>SMARCB1</i> | 2 | Soft Tissue 20%; CNS 7% | Diagnostic for AT/RT |
| <i>SMO</i> | 1 | Skin 3%; Colorectal 3% | Clinical Studies |
| <i>SRC</i> | 1 | Skin 3% | Clinical Studies |
| <i>STK11</i> | 1 | Lung 5%; GI 20% | Clinical Studies |
| <i>TP53</i> | 33 | | Clinical Studies |
| <i>VHL</i> | 6 | Kidney 36% | Clinical Studies |

- 1) *ALK* gene fusions are not detected by this assay
2) *ERBB2* amplification is not detected by this assay